


## PCT

WIPO

PCT

## INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Article 36 and Rule 70)

|  |  |   |   |
|--|--|---|---|
| Applicant's or agent's file reference<br>160721  |  | <b>FOR FURTHER ACTION</b>   | See Notification of Transmittal of International Preliminary Examination Report (Form PCT/IPEA/416) |
| International application No.<br>PCT/NL99/00737  | International filing date (day/month/year)<br>02/12/1999 | Priority date (day/month/year)<br>02/12/1998                            |   |
| International Patent Classification (IPC) or national classification and IPC<br>C12N15/53  |  |   |   |
| Applicant<br>CENTRUM VOOR PLANTENVEREDELINGS EN REPRODUKTIE ...  |  |   |   |
| <p>1. This international preliminary examination report has been prepared by this International Preliminary Examining Authority and is transmitted to the applicant according to Article 36.</p> <p>2. This REPORT consists of a total of 9 sheets, including this cover sheet.</p> <p><input type="checkbox"/> This report is also accompanied by ANNEXES, i.e. sheets of the description, claims and/or drawings which have been amended and are the basis for this report and/or sheets containing rectifications made before this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions under the PCT).</p> <p>These annexes consist of a total of sheets.</p>  |  |   |   |
| <p>3. This report contains indications relating to the following items:</p> <ul style="list-style-type: none"><li>I <input checked="" type="checkbox"/> Basis of the report</li><li>II <input type="checkbox"/> Priority</li><li>III <input type="checkbox"/> Non-establishment of opinion with regard to novelty, inventive step and industrial applicability</li><li>IV <input checked="" type="checkbox"/> Lack of unity of invention</li><li>V <input checked="" type="checkbox"/> Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement</li><li>VI <input type="checkbox"/> Certain documents cited</li><li>VII <input checked="" type="checkbox"/> Certain defects in the international application</li><li>VIII <input checked="" type="checkbox"/> Certain observations on the international application</li></ul> |  |   |   |
| Date of submission of the demand<br><br>03/07/2000   |  | Date of completion of this report<br><br>22.03.2001                     |   |
| Name and mailing address of the international preliminary examining authority:<br><br> European Patent Office<br>D-80298 Munich<br>Tel. +49 89 2399 - 0 Tx: 523656 epmu d<br>Fax: +49 89 2399 - 4465  |  | Authorized officer<br><br>Kurz, B<br><br>Telephone No. +49 89 2399 7319 |   |



# INTERNATIONAL PRELIMINARY EXAMINATION REPORT

International application No. PCT/NL99/00737

## I. Basis of the report

1. This report has been drawn on the basis of *(substitute sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to the report since they do not contain amendments (Rules 70.16 and 70.17).):*

### Description, pages:

1-123 as originally filed

### Claims, No.:

1-52 as originally filed

### Drawings, sheets:

1/18-18/18 as originally filed

### Sequence listing part of the description, pages:

1-93, filed with the letter of 15.2.00

2. With regard to the **language**, all the elements marked above were available or furnished to this Authority in the language in which the international application was filed, unless otherwise indicated under this item.

These elements were available or furnished to this Authority in the following language: , which is:

- ☐ the language of a translation furnished for the purposes of the international search (under Rule 23.1(b)).
- ☐ the language of publication of the international application (under Rule 48.3(b)).
- ☐ the language of a translation furnished for the purposes of international preliminary examination (under Rule 55.2 and/or 55.3).

3. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, the international preliminary examination was carried out on the basis of the sequence listing:

- ☐ contained in the international application in written form.
- ☐ filed together with the international application in computer readable form.
- ☐ furnished subsequently to this Authority in written form.
- ☒ furnished subsequently to this Authority in computer readable form.
- ☐ The statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.
- ☐ The statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished.

4. The amendments have resulted in the cancellation of:

# INTERNATIONAL PRELIMINARY EXAMINATION REPORT

International application No. PCT/NL99/00737

- ☐ the description, pages:
- ☐ the claims, Nos.:
- ☐ the drawings, sheets:

5. ☐ This report has been established as if (some of) the amendments had not been made, since they have been considered to go beyond the disclosure as filed (Rule 70.2(c)):

*(Any replacement sheet containing such amendments must be referred to under item 1 and annexed to this report.)*

6. Additional observations, if necessary:

## IV. Lack of unity of invention

1. In response to the invitation to restrict or pay additional fees the applicant has:

- ☐ restricted the claims.
- ☐ paid additional fees.
- ☐ paid additional fees under protest.
- ☒ neither restricted nor paid additional fees.

2. ☐ This Authority found that the requirement of unity of invention is not complied and chose, according to Rule 68.1, not to invite the applicant to restrict or pay additional fees.

3. This Authority considers that the requirement of unity of invention in accordance with Rules 13.1, 13.2 and 13.3 is

- ☐ complied with.
- ☒ not complied with for the following reasons:  
**see separate sheet**

4. Consequently, the following parts of the international application were the subject of international preliminary examination in establishing this report:

- ☐ all parts.
- ☒ the parts relating to claims Nos. 1-16 and 49 (completely); 50-52 (partially).

## V. Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

|             |      |        |                          |
|-------------|------|--------|--------------------------|
| Novelty (N) | Yes: | Claims | 12, 13, 16               |
|             | No:  | Claims | 1-11, 14, 15, 49, 51, 52 |

# INTERNATIONAL PRELIMINARY EXAMINATION REPORT

International application No. PCT/NL99/00737

|                               |      |        |                     |
|-------------------------------|------|--------|---------------------|
| Inventive step (IS)           | Yes: | Claims | 12, 13, 16          |
|                               | No:  | Claims | 1-11, 14, 15, 49-52 |
| Industrial applicability (IA) | Yes: | Claims | 1-16, 49-52         |
|                               | No:  | Claims | -                   |

2. Citations and explanations  
**see separate sheet**

## VII. Certain defects in the international application

The following defects in the form or contents of the international application have been noted:  
**see separate sheet**

## VIII. Certain observations on the international application

The following observations on the clarity of the claims, description, and drawings or on the question whether the claims are fully supported by the description, are made:  
**see separate sheet**

**Re Item I**

**Basis of the opinion**

1. Examination was based on the sequence listing filed with the letter of 15.2.2000 in computer readable form.  
Whether the amendments filed with the same letter are allowable will be discussed in the European phase as the sequences concerned have not been examined at the present stage due to lack of unity (see Item IV).
2. The applicant is informed that according to Rule 66.8 PCT amendments can only be filed in the form of replacement sheets. As the examiner himself cannot file replacement sheets, the amendments proposed in the response to the Written Opinion were not taken into consideration. Examination was carried out based on the claims as originally filed.

**Re Item IV**

**Lack of unity of invention**

In contrast to the findings of the ISA, the IPEA is of the opinion that the present application lacks unity as required by Article 3(4)(iii) PCT and Rule 13 PCT:

Rule 13.1 states that for unity of invention to be present, all subject-matter should be linked by a single general inventive concept.

The present application pertains to numerous nucleotide sequences coding for the following proteins: strawberry alcohol acyl transferase, strawberry aminotransferase, strawberry thiolase, strawberry pyruvate decarboxylase, strawberry alcohol dehydrogenase, and mango esterase. In addition, various methods for altering the level of volatile esters in fruits or the production of volatile esters, as well as antibodies against the claimed proteins and diagnostic kits are claimed.

All the sequences claimed represent solutions to **different technical problems**, and consequently no common concept linking the different groups can be identified. The assumption that all the claimed protein sequences -amongst other functions- may take part in the production of esters in fruits is not considered a common concept.

Furthermore, since also **no special technical or functional feature** (Rule 13.2 PCT)

**INTERNATIONAL PRELIMINARY  
EXAMINATION REPORT - SEPARATE SHEET**

---

International application No. PCT/NL99/00737

could be identified to provide a linking concept between the different groups of the invention, each of these groups must be regarded as a separate invention (N.B. The use of the term "invention" here in no way implies recognition of an inventive step for the subject-matter of any group).

The requisite unity of invention therefore does not exist.

The present application contains the following six groups:

Group 1: Claims 1-16 and 49 (completely); claims 50-52 (partially)

DNA and amino acid sequences of strawberry and lemon alcohol acyl transferase, vectors containing them, antibodies, methods of regulating ester production by using them, and methods for producing esters in microorganisms, diagnostic kits and the use thereof

Group 2: Claims 17-19 (completely); claims 43, 44, 47, 48, 50-52 (partially)

DNA and amino acid sequences of strawberry aminotransferase, modified plants, antibodies, methods for regulating ester formation, methods for producing esters in microorganisms, diagnostic kits and the use thereof

Group 3: Claims 20-22 (completely); claims 43, 44, 48 and 50-52 (partially)

DNA and amino acid sequences of strawberry thiolase, modified plants, antibodies, methods for regulating ester formation, methods for producing esters in microorganisms, diagnostic kits and the use thereof

Group 4: Claims 23-25 (completely); claims 43, 44, 46-48, and 50-52 (partially)

DNA and amino acid sequences of strawberry pyruvate decarboxylase, modified plants, antibodies, methods for regulating ester formation, methods for producing esters in microorganisms, diagnostic kits and the use thereof

**INTERNATIONAL PRELIMINARY  
EXAMINATION REPORT - SEPARATE SHEET**

---

International application No. PCT/NL99/00737

Group 5: Claims 26-39 (completely); claims 43--48 and 50-52 (partially)

DNA and amino acid sequences of strawberry alcohol dehydrogenase, modified plants, antibodies, methods for regulating ester formation, methods for producing esters in microorganisms, diagnostic kits and the use thereof

Group 6: Claims 40-42 (completely); claims 43-48, and 50-52 (partially)

DNA and amino acid sequences of mango esterase, modified plants, antibodies, methods for regulating ester formation, methods for producing esters in microorganisms, diagnostic kits and the use thereof

**As the applicant did not reply to the invitation to restrict or to pay additional fees, group 1 as indicated above and in the invitation was examined.**

The applicant is informed that during the course of the proceedings further objections concerning unity of invention (Article 3(4)(iii) PCT and Rule 13 PCT) might be raised.

**Re Item V**

**Reasoned statement under Rule 66.2(a)(ii) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement**

The present application pertains to sequences encoding strawberry and lemon alcohol acyl transferases as well as to vectors encoding them. Claimed are also methods of producing the respective enzymes in microorganisms.

Reference is made to the following documents:

- D1: MANNING K.: 'Isolation of a set of ripening-related genes from strawberry: their identification and possible relationship to fruit quality traits' PLANTA, vol. 205, 1998, pages 622-631
- D2: WO 97 27295 A (HORTICULTURE RESEARCH INTERNAT; MANNING KENNETH (GB)) 31 July 1997 (1997-07-31) cited in the application

**1. Novelty (Article 33(2) PCT):**

Claims 1-11, 14, and 49 are considered not to be novel for the following reasons:

- 1.1 In table 1, page 624, document D1 discloses a clone designated FAN R48 with the database accession number AF041395. The date of the database entry is 08 July 1998. This clone is 98% identical to Seq. ID No. 1 over 566 nucleotides. In view of this document, claims 1-4 and 7-9 cannot be considered novel.

The function as well as the polypeptide sequence of a known nucleotide sequence are inherent features of this sequence. Consequently, claims making reference to or encompassing this known sequence have to be regarded as not novel as long as the known parts/sequences are not explicitly disclaimed.

- 1.2 Document D2 discloses sequences related to the ripening of strawberry. One of these (partial) sequences designated StrawRipe C (see page 19 Table 1) is identical to part of Seq. ID. No. 1 of the present application. Vectors and plants comprising the respective sequences are claimed in D2, too. With respect to D2 thus claims 1-5, 7-11, 14, 15, and 49 are not novel.
- 1.3 As neither the nucleotide nor the polypeptide sequences at present are considered novel over the art, also a diagnostic kit comprising such sequences (claim 51) and the use of this kit (claim 52) cannot be considered novel.
- 1.4 Due to a lack of clarity (see Item VIII) also claim 6 at present is not considered as novel as no function of the claimed sequence is given.

**2. Inventive Step (Article 33(3) PCT):**

- 2.1 Documents D1 and D2 disclose sequences related to fruit ripening. Both documents do not give details about (putative) functions of those sequences which are relevant for the present application.
- However, it can be stated that both sequences enable the skilled person to isolate the sequences claimed in the present application in claims 1-11. These claims can consequently not be considered inventive.



**INTERNATIONAL PRELIMINARY  
EXAMINATION REPORT - SEPARATE SHEET**

---

International application No. PCT/NL99/00737

Furthermore, the knowledge of the DNA and the respective amino acid sequence enables the skilled person to produce antibodies by routine procedures (i.e. without any inventive activity). In view of the cited documents thus claim 50 also cannot be considered inventive.

**Re Item VII**

**Certain defects in the international application**

1. The reference contained in claim 51. c) seems to be incorrect as claim 47 is not related to antibodies.

**Re Item VIII**

**Certain observations on the international application**

1. Claims 2, 3, 4c, 5, 6c, 8, and 9 are unclear (Article 6 PCT) as neither the size nor the function of the fragments claimed are defined in any way. The present definition includes fragments of only a few nucleotides/amino acids which render the claims not novel (see Item V). Furthermore, no function of the DNA/amino acid sequences showing a certain degree of homology is given. Thus these sequences might code for or represent completely unrelated proteins. E.g. claim 2 encompasses a fragment of the DNA sequence of claim 1. In claim 1 the function of the claimed fragment is given, but in claim 2 no function is given for a "fragment of the fragment" of claim 1.

Claim 4 specifically relates to a sequence which has at least 25% homology. However, a sequence similarity or identity of 25% does not seem to be sufficient to characterise a DNA sequence unambiguously. In the description of the present application no evidence is given that sequences exist which have only 25% similarity and exhibit the same function.